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Genetic Stock Identification of Chinook Salmon Harvest on the Yukon River in 2004

Final Report for Project USRM 03-05 United States Yukon River Research and Management Fund

by

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Alaska Department of Fish and Game

Division of Commercial Fisheries



Symbols and Abbreviations

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Weights and measures (metric)		General		Measures (fisheries)	
centimeter	cm	Alaska Administrative		fork length	FL
deciliter	dL	Code	AAC	mideye-to-fork	MEF
gram	g	all commonly accepted		mideye-to-tail-fork	METF
hectare	ha	abbreviations	e.g., Mr., Mrs.,	standard length	SL
kilogram	kg		AM, PM, etc.	total length	TL
kilometer	km	all commonly accepted		Ç	
liter	L	professional titles	e.g., Dr., Ph.D.,	Mathematics, statistics	
meter	m		R.N., etc.	all standard mathematical	
milliliter	mL	at	@	signs, symbols and	
millimeter	mm	compass directions:		abbreviations	
		east	E	alternate hypothesis	H_A
Weights and measures (English)		north	N	base of natural logarithm	e
cubic feet per second	ft ³ /s	south	S	catch per unit effort	CPUE
foot	ft	west	W	coefficient of variation	CV
gallon	gal	copyright	©	common test statistics	$(F, t, \chi^2, etc.)$
inch	in	corporate suffixes:		confidence interval	CI
mile	mi	Company	Co.	correlation coefficient	
nautical mile	nmi	Corporation	Corp.	(multiple)	R
ounce	OZ	Incorporated	Inc.	correlation coefficient	
pound	lb	Limited	Ltd.	(simple)	r
quart	qt	District of Columbia	D.C.	covariance	cov
yard	yd	et alii (and others)	et al.	degree (angular)	0
·	•	et cetera (and so forth)	etc.	degrees of freedom	df
Time and temperature		exempli gratia		expected value	E
day	d	(for example)	e.g.	greater than	>
degrees Celsius	°C	Federal Information		greater than or equal to	≥
degrees Fahrenheit	°F	Code	FIC	harvest per unit effort	HPUE
degrees kelvin	K	id est (that is)	i.e.	less than	<
hour	h	latitude or longitude	lat. or long.	less than or equal to	≤
minute	min	monetary symbols		logarithm (natural)	ln
second	S	(U.S.)	\$, ¢	logarithm (base 10)	log
		months (tables and		logarithm (specify base)	log _{2,} etc.
Physics and chemistry		figures): first three		minute (angular)	'
all atomic symbols		letters	Jan,,Dec	not significant	NS
alternating current	AC	registered trademark	®	null hypothesis	H_{O}
ampere	A	trademark	TM	percent	%
calorie	cal	United States		probability	P
direct current	DC	(adjective)	U.S.	probability of a type I error	
hertz	Hz	United States of		(rejection of the null	
horsepower	hp	America (noun)	USA	hypothesis when true)	α
hydrogen ion activity	pН	U.S.C.	United States	probability of a type II error	
(negative log of)			Code	(acceptance of the null	
parts per million	ppm	U.S. state	use two-letter	hypothesis when false)	β
parts per thousand	ppt,		abbreviations	second (angular)	"
	‰		(e.g., AK, WA)	standard deviation	SD
volts	V			standard error	SE
watts	W			variance	
				population	Var
				sample	var

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ABSTRACT

Significant genetic variation exists among populations of Chinook salmon from the Yukon River drainage. Use of this variation for providing estimates of stock composition of fishery harvests has been possible since 1992. In 2004, a baseline of 17 SNP loci was used to estimate the stock composition of Chinook salmon harvests in the U.S. portion of the Yukon River. Of the samples collected from the subsistence fishery in District Y-1 and commercial fisheries in Districts Y-1, Y-2, and Y-5, approximately 4700 individuals were assayed for genetic variation at the 17 loci. Mixed stock analysis of these samples enabled the estimation of the stock composition of the harvest at three hierarchical levels: Country (U.S. and Canada), Broad-scale (Lower Yukon, Middle Yukon, and Canada), and Fine-scale (Lower Yukon, Middle Yukon, Canada Border, Pelly, Tatchun, Takhini, and Whitehorse). In District Y-1 the portion of harvest attributable to Canadian origin fish was consistently near 50% with the exception of two commercial fishing periods, when it dropped below 40%. In the District Y-2 harvest, Canadian stocks contributed to approximately half of the harvest over the four commercial fishing periods. In general, less than half of the five-year old and more than half of the six-year old Chinook salmon harvested were of Canadian origin.

Key words: Yukon River, Chinook salmon, genetic stock identification, SNP, commercial fishery, subsistence fishery.

INTRODUCTION

Chinook salmon *Oncorhynchus tshawytscha* stocks return to the mouth of the Yukon River in early June to spawn throughout the drainage. The upriver migration of some stocks traverses the border into Canada where spawning sites are located in tributaries in the Yukon Territory and British Columbia. These salmon stocks, in both U.S. and Canadian portions of the drainage, are managed to reach the necessary escapement goals (ADF&G 2004). Knowledge of the origin of Chinook salmon stocks harvested in the subsistence and commercial fisheries on the Yukon River is important for the successful management of these fisheries. The proportion of Canadian-origin Chinook salmon in fishery harvests in the U.S. waters of the Yukon River is necessary information for meeting the obligations of the Yukon River Salmon Agreement between the United States and Canada. Until 2004, the methodology used to estimate stock composition was scale pattern analysis. Recently developed genetic techniques and the collection of baseline data for Chinook salmon populations in the Yukon River drainage (Smith et al. 2005) demonstrated the increased power and resolution available from a genetic approach to estimate stock proportions in the harvest.

Early surveys of genetic variation among Chinook salmon populations in the Yukon River demonstrated significant genetic variation based on protein or allozyme variation (Gharrett et al. 1987; Beacham et al. 1989). While these studies discussed the potential use of this information for management of the resource, it was not until 1992 that a baseline of genetic information was completed and used for genetic stock identification (Wilmot et al. 1992; Templin et al. 2005). In 1997 the U.S. and Canada Joint Technical Committee (JTC 1997) reviewed stock identification techniques. Subsequently Alaska Department of Fish and Game (ADF&G) decided that scale pattern analysis provided sufficient stock-specific information for management applications. In 2002, there was renewed interest in genetic stock identification, and a joint collaboration began among ADFG, U.S. Fish and Wildlife Service (USFWS) and Canadian Department of Fisheries and Oceans (CDFO) to collect and develop DNA baselines for Yukon River Chinook salmon.

Two types of DNA markers, SNPs (Smith et al. 2005) and microsatellites are being explored to provide a replacement for the allozyme baseline. Under standards set for DNA-based technologies by the Joint Technical Committee of the Yukon River Salmon Treaty, both ADFG and CDFO are surveying a standardized set of loci in Yukon River Chinook salmon populations. While a transparent, fully sharable baseline based on microsatellites and SNPs should be

available for use in the near future, in 2004 only the SNP baseline was standardized and could be used to provide estimates from that year's fishery samples.

This project was developed to use mixed stock analysis on SNP data to estimate the contribution of the three reporting groups (Lower Yukon, Middle Yukon, and Canada) to the harvest of Chinook salmon in commercial and subsistence fisheries in the U.S. portion of the Yukon River drainage. This baseline was recently augmented from the original 10 SNP loci by the addition of nine new SNP loci. The effect of adding these new loci will be investigated as part of this analysis. The methods employed to provide estimates were determined so that the accuracy and precision of the estimates would be within 5% of the true value 90% of the time.

OBJECTIVES

The goal of this project is to estimate the stock composition of the harvests of Chinook salmon in selected commercial and subsistence fisheries on the Yukon River in 2004. The fisheries to be sampled and analyzed are:

- 1) District Y-1 subsistence fishery,
- 2) District Y-1 commercial fishery periods 1-8,
- 3) District Y-2 commercial fishery periods 1-4,
- 4) District Y-4 subsistence fishery, and
- 5) District Y-5 commercial fishery.

METHODS

SAMPLE COLLECTION

The set of collections from Chinook salmon populations that comprise the baseline (Table 1) was assembled as a part of a three-laboratory collaboration (ADF&G, CDFO, and USFWS) to survey microsatellite variation in the Yukon River drainage. Genetic material from each population was distributed among the laboratories in the form of tissues or DNA extract. Further discussion of the baseline is described in Smith et al. (2005).

During 2004, Chinook salmon were sampled from the commercial, subsistence, and test fisheries in the U.S. portion of the river (Table 2). Samples were collected randomly during each fishing period, with a target sample size of 400 individuals, while sampling the harvest for age, sex, and length (ASL) data. In general, the tissues collected were axillary processes preserved in ethanol. When that tissue was not available, scales collected during ASL sampling of the harvest were used. Two scales from each individual (three were collected) were removed from the scale card after it was pressed and ASL analyses had been completed.

LABORATORY METHODS

Individuals were assayed for their genotypes at nine SNPs and combined with the previously existing baseline of 10 SNP loci (Table 3). SNP genotyping was performed in 384-well reaction plates, with two wells in each plate as negative controls (no-template) and two wells as positive controls (one for each allele). Each polymerase chain reaction (PCR) was conducted in a 5μ L

volume consisting of $0.10\mu L$ template DNA in 1X TaqMan Universal Buffer (ABI), 900nM each PCR primer, and 200nM each probe. Thermal cycling was performed on a Dual 384-Well GeneAmp PCR System 9700 as follows: an initial denaturizing step of 10 min at 95°C followed by 50 cycles of: 92°C for 15 sec and annealing/extension temperature for 1 or 1.5 min. Cycling was conducted at a ramp speed of 1°C per second. The plates were read on an ABI PRISM 7900HT Sequence Detection System after amplification and scored using Sequence Detection Software 2.2 (ABI).

The SNP data collected were individual diploid genotypes for each locus. Genotype data were stored as output text files on a network drive. The data on this network are backed up nightly. Long term storage of the data is in an Oracle database, *LOKI*, supported and maintained by ADF&G.

QUALITY CONTROL METHODS

Several measures were implemented to insure the quality of data produced:

- 1) Sample sheets which contain information for each plate of extracted DNA in the lab were created in a standard format. Once DNA was extracted or obtained from an outside source, an Excel file was created containing sample information for each well on that plate. This sample sheet followed the plate through all phases of a project, minimizing the risk of misidentification of samples through human induced errors.
- 2) Genotypes were assigned to individuals using a double-scoring system. Two observers independently produced allele scores for an entire project before the two data sets were compared. Discrepancies between the two sets of scores were then resolved with one of three possible outcomes: 1) one score was accepted and the other rejected, 2) both scores were rejected and the score is blanked, or 3) the sample was rerun.
- 3) Approximately eight percent of the individuals, eight samples from each 96 well DNA extraction plate, were reanalyzed for all loci. This insured that the data are reproducible and any errors created from the processing of individual plates were corrected.

STATISTICAL ANALYSIS

Individual genotype data were summarized as allele frequencies for all SNP loci in each population. Estimates of the population frequency of individual alleles for each locus were calculated from the observed frequency of the allele in the representative sample. Observed and expected heterozygosity (HWE) and conformation of genotype frequencies to HWE expected ratios was assessed using the exact test in Goudet (1995). Two measures of population subdivision were calculated from allele frequency differences: Cavalli-Sforza and Edwards' chord distances (*PHYLIP*, version 3.6; Felsenstein 2005) and F_{ST} (Weir and Cockerham 1984). *GENEPOP* (Raymond and Rousset 1995) was used to calculate F_{ST} values. Population structure was visualized as a tree (unweighted pair-group method; Sneath and Sokal 1973) to view genetic similarities between populations reflected in the interpopulation chord distances.

MIXED STOCK ANALYSIS

Simulations

Simulations were conducted to evaluate the potential application of genetic stock identification to mixtures of Chinook salmon harvested in Yukon River fisheries using the same methods

reported previously (Smith et al. 2005). These simulations were used to help assess whether the baseline of allele frequencies at the SNP markers provides sufficient information to identify individual stocks or groups of stocks (reporting groups) in hypothetical mixtures.

Reporting groups for genetic stock identification of Yukon River Chinook salmon have already been defined based on a combination of genetic similarity, geographic features, and management applications (Lower Yukon, Middle Yukon and Canada). Simulations were performed using the Statistical Package for Analyzing Mixtures (SPAM version 3.6; Debevec et al. 2000). Baseline and mixture genotypes were randomly generated from the baseline allele frequencies assuming Hardy-Weinberg equilibrium. Each simulated mixture (N = 400) was composed 100% of the stock or reporting group under study. When a reporting group mixture was simulated, all stocks in the stock group contributed equally to the mixture. Average estimates of mixture proportions and 90% confidence intervals were derived from 1000 simulations. Reporting groups with mean correct estimates of 90% or better are considered highly identifiable in fishery applications. Reporting groups with mean correct estimates lower than 90% can still be considered identifiable in mixtures, but sources of misallocation should be considered when interpreting the results.

Mixture analysis

Stock composition estimates for the three stock groups (Lower Yukon, Middle Yukon, and Canada) were generated using SPAM version 3.7 (Debevec et al. 2000). For each estimation procedure, genotypes were removed from the estimation procedure if their probability of occurring was near zero. The mixture estimates have an unknown group containing the percent of the mixture that was removed. Further, we deleted any individual missing data at four or more loci. Individual population or stock estimates were first calculated, and then summed into reporting regions. Ninety percent confidence intervals for all group contribution estimates were computed from 1000 bootstrap resamples of the baseline and mixture genotypes. For each resample, contribution estimates were generated for all populations and summed to the group level. The 1000 estimates for a group were then sorted from lowest to highest with the 51st and 950th values in the sequence taken respectively as the lower and upper bounds of the 90% confidence interval for that group.

To investigate the precision and accuracy of stock composition estimates from non-simulated mixtures, three mixtures of known composition were analyzed using this baseline. To create the mixtures, the genotypes of 95 individuals were randomly selected and removed from the samples available for the Andreafsky, Chena, and Tatchun rivers. Baseline allele frequencies were recalculated for these three populations and a revised baseline was created. Three mixtures were formed using only the selected individuals from each population, creating mixtures analogous to the 100% simulated mixtures. Correct allocation to population and reporting group (fine-scale and country) was measured as described above.

The stock composition of the commercial and subsistence fishery harvests of Chinook salmon in the Yukon River were estimated from the genotypes of the individuals sampled during each period and location. Estimates of the age-structured harvest components were also available. Genetic tissue samples were collected as part of the ASL sampling of the harvests, and, once the scales had been aged, the individuals in each sample could be separated into the following groups, 1.2, 1.3, 1.4, and 1.5 (4 - 7 years old, respectively). Adequate sample sizes for five- and six-year olds were available for further independent analyses. Mixed stock analysis was performed on the subsets of individuals (by age) following the procedure described above.

RESULTS

BASELINE SAMPLES

A total of 3,236 individuals from 34 collections representing 23 populations were analyzed (Table 1) to create the baseline. Allele frequencies at one SNP locus (*Ots_GnRH-271*) were fixed for a single allele in all baseline collections and this locus was dropped from the analysis.

During 2004, 5,707 Chinook salmon were sampled as part of 16 collections from the commercial and subsistence fishery harvests in the U.S. portion of the Yukon River drainage (Table 2). Of these, a total of 4,673 individuals were analyzed for allelic variation at 17 SNP loci. Because it is more efficient to analyze sets of 95 individuals (rather than 100) in the laboratory, in many cases only 380 of the 400 individuals available were analyzed. Sampling theory (Thompson 1987) shows that this reduction in sample size should have little effect on the precision or accuracy of the estimate. The quality control checks employed demonstrated an error rate of 0.04%.

Samples taken from the commercial fishery in District Y-6 were not analyzed, because no stock composition estimates were considered necessary for this Tanana River-only fishery harvest.

STATISTICAL ANALYSIS

Collections taken in multiple years from the same location were pooled for further analyses. After adjusting for the number of tests, no significant differences were found between the temporally spaced collections from the same location.

Genetic distances were calculated between each pair of populations and the unweighted pair-group method with arithmetic mean (UPGMA) was used to create a dendrogram of genetic relationships between the populations in the baseline (Figure 2). The greatest distinction identified in this analysis is between Chinook salmon populations of U.S. and Canadian origins. Within the U.S. populations, two clusters were formed. The first contained populations from the lower portion of the Yukon and Koyukuk river drainages and the second group contained populations from the upper U.S. portion of the Yukon River and the Tanana and Porcupine rivers. Within the main Canadian cluster, populations also grouped geographically into five smaller regional clusters: populations near the U.S./Canada border, the Pelly River drainage, Takhini River drainage, populations from the Tatchun area, and the Whitehorse Hatchery collection.

MIXED STOCK ANALYSIS

Simulations

Reporting groups for mixed stock analysis of Chinook salmon in the Yukon River were defined based on previous studies (Templin et al. 2005; Smith et al. 2005) and supported by the structure revealed in this analysis: 1) Lower Yukon – Andreafsky River, Anvik River, Tozitna River, and Gisasa River, 2) Middle Yukon – Henshaw Creek, South Fork Koyukuk River, Chena River, Salcha River, Beaver Creek and Chandalar River, 3) Canada Border – Chandindu River, and Klondike River, 4) Pelly – Pelly River, Mayo River, Stewart River, and Blind Creek, 5) Tatchun – Tatchun River, Nisutlin River, Nordenskiold River, and Big Salmon River, 6) Takhini – Takhini River and Stoney Creek, and 7) Whitehorse Hatchery. Simulation studies based on this structure indicate that these reporting groups are highly identifiable in mixtures. When simulated mixtures composed entirely from a single reporting group were treated as mixtures of unknown

origin more than 90% of the mixture was correctly identified to region-of-origin (Table 4). As expected, a higher level of distinction was also seen when simulating mixtures from each nation (98% correct allocation).

Mixture Analysis

When mixtures of known composition were created from each of the Andreafsky, Chena, and Tatchun rivers, estimates were highly accurate (>90%) when identifying the country and fine-scale region of origin (Table 5). Correct identification to population was less accurate, ranging from 51% to 87%.

Estimates of stock composition in the commercial harvest in District Y-1 of the Yukon River indicate that Chinook salmon of Canadian origin contributed more than 50% of the harvest during three of the six commercial fishing periods (Table 6; Figure 3). Most of these Canadian salmon were estimated to be from the Pelly River and Tatchun regions followed by contributions from the Canada Border region stocks. During periods 3 (July 24-25) and 6 (July 2-3) the proportion of Canadian populations in the harvest dropped to 37% and 25%, respectively. These reductions were matched by an increase in the presence of Lower Yukon populations in the harvest (48% and 55% respectively).

Stock composition estimates of the Canadian contribution to the commercial harvest in District Y-2 varied more widely over the two weeks of the fishery (Table 7; Figure 4). The Canadian component of the harvest was approximately 50% over the whole period, but peaked at 61% (June 20) and dropped to 36% at the end (June 27). The Middle Yukon portion of the harvest dropped over the course of the four periods from 41% to 16%, while the Lower Yukon contribution increased from 8% to 49%.

Approximately half of the harvest in the subsistence fishery in District Y-1 was estimated to be from each nation (Table 8). The estimated contribution of Canadian populations to the subsistence harvest in District Y-4 was 59%, most of which came from the Tatchun stock group. Middle Yukon populations made up almost the entire U.S. component of the harvest. The stock composition of the subsistence harvest in District Y-5 was even more extreme; Canadian populations contributed 85% of the harvest. Again, Middle Yukon populations contributed almost the entire U.S. portion of the harvest.

Stock composition was also estimated independently by age class. Sufficient samples were available to estimate the composition of the five- and six-year old components for the Y-1, Y-2, and Y-5 commercial fisheries and the Y-1 and Y-4 subsistence fisheries (Appendix A; Appendix B; Figures 5 and 6). Sample sizes for the five-year old components were low ranging from 48 to 119; these lower sample sizes result in lower precision (larger 90% confidence intervals). Samples sizes for the six year old components were uniformly larger ranging from 158 to 293.

DISCUSSION

The baseline used for mixed stock analysis of Chinook salmon harvests in the Yukon River has been extended by the work done in this project. Allelic frequencies at eight additional SNP loci were added to the baseline and the additional information increased the precision and accuracy provided by the baseline for mixed stock analysis. The correct allocation to each reporting group was increased by a few percentage points over the previous baseline with only nine SNP loci (Smith et al. 2005b) and all reporting groups (including Pelly and Whitehorse) are now above the

90% threshold. When combined with the results of the known mixture samples, this baseline of SNP loci has been demonstrated to be sufficiently accurate and precise for estimating the stock composition of fishery harvests.

In 2004, the relative proportion of Middle Yukon populations in the District Y-1 harvest was greatest during the early part of the season (the subsistence fishery and the first period of the commercial fishery) after which, these populations contributed at a consistently reduced level (15%-20%). A similar pattern was observed in the District Y-2 commercial fishery. The relative contribution of Canada populations to the harvest in both districts Y-1 and Y-2 fluctuated around 50% (38%-61%) throughout the season before dropping to 25% during Period 6 in Y-1. The Lower Yukon group contributed the least early in the season (8%) before climbing to contribute approximately half the harvest during the last periods in both districts.

A consistent signal is observed in the relative stock proportions when estimates of the stock composition of the fisheries in each of districts Y-1 and Y-2 are compared for closely matching dates (Y-2 lagged by one period from Y-1). For example, the stock composition of the Period 2 (June 20) harvest in Y-2 is very similar to the composition of the Period 1 (June 15-18) harvest in Y-1. This is also reflected in the estimates from five- and six-year old portions of the harvest, where larger proportions of the harvest at these ages were of U.S.-origin.

In 2004, Chinook salmon from the U.S. appear to be more prevalent in the five-year old age class than the six-year old age class. In districts Y-1 and Y-2, the U.S. component was consistently larger for five-year olds than six-year olds with the exception of Period 4 in Y-1 and Period 2 in Y-2.

In the District Y-4 subsistence fishery and the District Y-5 commercial fishery the Canada stock group contributed the largest part of the harvest (58% and 85% respectively). As might be expected due to these districts being located further upriver, almost the entire U.S. component of the harvest was from the Middle Yukon stock group.

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TABLES AND FIGURES

Table 1.—Chinook salmon collections from the Yukon River drainage organized hierarchically into reporting groups for mixed stock analysis.

]	Reporting Gro	up				
Country	Broad scale	Fine scale	Location	Sample size	Year collected	Agency
United Sta						
	Lower Yukor	1				
		Lower Yuk	con			
			Anvik	85	2003	ADFG
			Andreafsky	208	2003	USFWS
			Tozitna	250	2003	USFWS
			Gisasa	228	2001	USFWS
	Middle Yuko	n				
		Middle Yuk	con			
			Henshaw	150	2001	USFWS
			S. Fork Koyukuk	56	2003	
			Chena	200	2001	USFWS
			Salcha	55	2003	USFWS
			Beaver	100	1997	USFWS
			Chandalar	117	2002, 2003	USFWS
Canada						
	Canada					
		Border				
			Chandindu	158	2001	CDFO
			Klondike	80	2001, 2003	CDFO
		Pelly				
			Blind	138	2003	CDFO
			Pelly	150	1996, 1997	CDFO
			Mayo	165	1992, 2003	CDFO
			Stewart	99	1997	CDFO
		Tatchun				
			Big Salmon	119	1987, 1997	CDFO
			Tatchun	285	1987, 1996, 1997	CDFO
			Nisutlin	56	1987, 1997	CDFO
			Nordenskiold	56	2003	CDFO
		Takhini				
			Takhini	168	1997, 2002, 2003	CDFO
			Stoney	185	1992	CDFO
		Whitehorse				
			Whitehorse	128	1985, 1997	CDFO

Note: The sample size, year collected and collecting agencies (ADF&G – Alaska Department of Fish and Game; USFWS – United States Fish and Wildlife Service; CDFO – Fisheries and Oceans Canada) are provided for each location.

Table 2.—Chinook salmon collections from selected commercial and subsistence fishery harvests in the Yukon River drainage, 2004.

			Samp	le size
District	Period	Location	Collected	Analyzed
Commercia	ıl			
Y1	1	Emmonak	400	380
	2	Emmonak	400	380
	3	Emmonak	400	380
	4	Emmonak	400	380
	5	Emmonak	400	380
	6	Emmonak	200	190
	7	Emmonak	190	0^1
	8	Emmonak	54	0^1
Y2	1	St. Marys	400	380
	2	St. Marys	400	380
	3	St. Marys	400	380
	4	St. Marys	400	380
Y5			480	380
Y6			480	0^{a}
		Total - commercial	5004	3990
Subsistence	2			
Y 1		Emmonak	400	380
Y4		Ruby/Galena	303	303
		Total - subsistence	703	683
	,	Гotal	5707	4673

^a Only scales were available, and DNA extracted was insufficient for analysis.

Table 3.–Single nucleotide polymorphism loci assayed in individuals from baseline and fishery collections.

Locus	Source	Range of Common Allele	$\mathbf{H}_{\mathbf{o}}$	$\mathbf{H}_{\mathbf{s}}$	$\mathbf{F}_{\mathbf{ST}}$
Ots_E2-275 †	Smith et al. 2005a	0.455-0.923	0.403	0.400	0.072
Ots_GH2	Smith et al. 2005a	0.129-0.783	0.413	0.437	0.130
Ots_GnRH-271 †	Smith et al. 2005a	1.000	-	-	-
Ots_IGF-I.1-76 †	Smith et al. 2005c	0.371-0.868	0.412	0.401	0.113
Ots_Ikaros-250 †	Smith et al. 2005c	0.791-1.000	0.152	0.154	0.033
Ots_ins-115 †	Smith et al. 2005c	0.973-1.000	0.004	0.004	0.018
Ots_LWSop-638 [†]	Smith et al. 2005c	0.897-1.000	0.045	0.047	0.032
Ots_MHC1	Smith et al. 2005a	0.325-0.797	0.452	0.455	0.047
Ots_MHC2	Smith et al. 2005a	0.840-1.000	0.056	0.058	0.055
Ots_Ots2	Smith et al. 2005a	0.859-1.000	0.089	0.097	0.034
Ots_P450	Smith et al. 2005a	0.513-0.875	0.379	0.379	0.032
Ots_P53	Smith et al. 2005a	0.221-0.720	0.475	0.468	0.069
Ots_Prl2	Smith et al. 2005a	0.291-1.000	0.405	0.413	0.109
Ots_SL	Smith et al. 2005a	0.714-0.986	0.232	0.233	0.053
Ots_SWS1op-182 †	Smith et al. 2005c	0.688-1.000	0.189	0.179	0.088
Ots_Tnsf	Smith et al. 2005a	0.450-0.890	0.310	0.339	0.097
Ots_u4-92 †	Smith et al. 2005c	0.930-1.000	0.017	0.016	0.038
Ots_u6-75 †	Smith et al. 2005c	0.510-0.840	0.415	0.427	0.043

Note: Loci marked with (†) were added to the original baseline described in Smith et al. (2005b)

Table 4.—Mean reporting group allocations of simulated mixtures of Yukon River Chinook salmon from the baseline of 17 SNP loci.

Region	P	90% CI
Reporting Groups		
Lower Yukon	0.974	(0.940 - 0.998)
Middle Yukon	0.969	(0.928 - 0.998)
Canada Border	0.940	(0.873 - 0.991)
Pelly	0.924	(0.847 - 0.985)
Tatchun	0.939	(0.862 - 0.993)
Takhini	0.962	(0.913-0.996)
Whitehorse	0.908	(0.821-0.980)
Country		
US	0.978	(0.948-1.000)
Canada	0.983	(0.951-1.000)

Note: Individual genotypes in the simulated mixtures were derived entirely from a single reporting group or country, therefore the expected value of the estimate is 1.0.

Table 5.—Mean reporting group allocations of three different mixtures (n = 95) of Yukon River Chinook salmon from three populations in the baseline.

Allocation to	Andı	eafsky River	C	hena River	Tatchun River		
Reporting Region	on P 90% CI		P	90% CI	P	90% CI	
Mixture Population	0.677		0.511		0.872		
Reporting Groups							
Lower Yukon	1.000	(0.900-1.000)	0.045	(0.000-0.133)	0.000	(0.000 - 0.028)	
Middle Yukon	0.000	(0.000 - 0.097)	0.939	(0.819-1.000)	0.018	(0.000 - 0.101)	
Canada Border	0.000	(0.000-0.000)	0.000	(0.000 - 0.024)	0.042	(0.000-0.135)	
Pelly	0.000	(0.000-0.000)	0.006	(0.000 - 0.084)	0.006	(0.000 - 0.150)	
Tatchun	0.000	(0.000-0.000)	0.011	(0.000 - 0.055)	0.934	(0.719 - 0.984)	
Takhini	0.000	(0.000-0.000)	0.000	(0.000-0.000)	0.000	(0.000 - 0.076)	
Whitehorse	0.000	(0.000-0.000)	0.000	(0.000-0.000)	0.000	(0.000 - 0.000)	
Country							
US	1.000	(0.946-1.000)	0.984	(0.752 - 1.000)	0.018	(0.000 - 0.110)	
Canada	0.000	(0.000 - 0.054)	0.016	(0.000 - 0.248)	0.982	(0.891-1.000)	

Note: Individuals were removed from the baseline samples, the baseline allele frequencies were recalculated, and the removed individuals were treated as single-population mixtures of known origin.

Table 6.—Estimated proportional stock composition (P) and 90% confidence intervals of Chinook salmon harvested from the commercial fishery in District Y-1 of the Yukon River, 2004.

	Period 1	Pe	eriod 2		Period 3]	Period 4		Period 5]	Period 6	
	June 15-18	June 20-21		June 24-25		June 27-28		July 1		July 2-3		
	N = 378	N = 379		N = 376			N = 379		N = 378		N=189	
P	90% CI	P	90% CI	P	90% CI	P	90% CI	P	90% CI	P	90% CI	
Reporting Region												
Lower Yukon 0.08	31 (0.047-0.133)	0.316 (0	0.253-0.367)	0.482	(0.402 - 0.539)	0.277	(0.211-0.329)	0.398	(0.339-0.469)	0.553	(0.454-0.665)	
Middle Yukon 0.34	45 (0.290-0.416)	0.183 (0	0.135-0.256)	0.145	(0.097-0.221)	0.181	(0.133-0.243)	0.164	(0.104-0.235)	0.195	(0.077-0.283)	
Canada Border 0.0	77 (0.000-0.134)	0.035 (0	0.000-0.095)	0.000	(0.000 - 0.039)	0.055	(0.006-0.106)	0.009	(0.000 - 0.056)	0.005	(0.000 - 0.068)	
Pelly 0.10	02 (0.039-0.216)	0.127 (0	0.046-0.199)	0.072	(0.012 - 0.149)	0.008	(0.000-0.103)	0.119	(0.023 - 0.192)	0.000	(0.000-0.102)	
Tatchun 0.2	70 (0.174-0.344)	0.246 (0	0.152-0.309)	0.203	(0.117-0.273)	0.326	(0.214-0.388)	0.234	(0.149-0.308)	0.218	(0.088 - 0.282)	
Takhini 0.03	57 (0.017-0.093)	0.054 (0	0.017-0.108)	0.076	(0.025 - 0.118)	0.081	(0.031-0.136)	0.076	(0.027-0.136)	0.030	(0.000-0.080)	
Whitehorse 0.00	68 (0.007-0.117)	0.039 (0	0.000-0.095)	0.023	(0.000-0.075)	0.071	(0.012-0.130)	0.000	(0.000-0.008)	0.000	(0.000-0.011)	
Country												
US 0.42	25 (0.372-0.504)	0.498 (0	0.441-0.572)	0.627	(0.565-0.692)	0.458	(0.393-0.522)	0.562	(0.502-0.638)	0.747	(0.647-0.817)	
Canada 0.5'	75 (0.496-0.628)	0.502 (0	0.429-0.559)	0.373	(0.309 - 0.435)	0.542	(0.479-0.607)	0.438	(0.362-0.499)	0.253	(0.183-0.353)	

Table 7.—Estimated proportional stock composition (P) and 90% confidence intervals of Chinook salmon harvested from the commercial fishery in District Y-2 of the Yukon River, 2004.

		Period 1		Period 2		Period 3		Period 4	
		June 15		June 20		June 24	June 27 N = 376		
		N = 378	N = 378			N = 378			
	P	90% CI	P	90% CI	P	90% CI	P	90% CI	
Reporting Region									
Lower Yukon	0.083	(0.033-0.137)	0.081	(0.042 - 0.133)	0.311	(0.240 - 0.362)	0.487	(0.423 - 0.552)	
Middle Yukon	0.414	(0.336 - 0.500)	0.306	(0.243 - 0.384)	0.178	(0.132 - 0.260)	0.156	(0.101 - 0.232)	
Canada Border	0.081	(0.017 - 0.160)	0.110	(0.032 - 0.176)	0.080	(0.020 - 0.138)	0.067	(0.000 - 0.114)	
Pelly	0.150	(0.064 - 0.257)	0.190	(0.099 - 0.288)	0.111	(0.037 - 0.185)	0.048	(0.004 - 0.134)	
Tatchun	0.260	(0.148 - 0.326)	0.297	(0.197 - 0.375)	0.288	(0.196 - 0.356)	0.187	(0.096 - 0.243)	
Takhini	0.012	(0.000 - 0.047)	0.008	(0.000-0.053)	0.022	(0.000 - 0.063)	0.019	(0.000 - 0.070)	
Whitehorse	0.000	(0.000-0.039)	0.007	(0.000-0.045)	0.010	(0.000-0.072)	0.036	(0.000-0.092)	
Country									
US	0.497	(0.423 - 0.572)	0.388	(0.329 - 0.461)	0.489	(0.432 - 0.558)	0.643	(0.590 - 0.713)	
Canada	0.503	(0.429-0.577)	0.612	(0.539-0.671)	0.511	(0.443 - 0.569)	0.357	(0.287-0.411)	

Table 8.—Estimated proportional stock composition (P) and 90% confidence intervals of Chinook salmon harvested from the subsistence fisheries in districts Y-1 and Y-4 and the commercial fishery in District Y-5 of the Yukon River, 2004.

	Di	istrict Y – 1	D	istrict Y – 4	Di	istrict Y – 5		
		N = 379		N = 283		N=375		
	P	90% CI	P	90% CI	P	90% CI		
Reporting Region								
Lower Yukon	0.124	(0.069 - 0.182)	0.030	(0.000 - 0.076)	0.015	(0.000 - 0.062)		
Middle Yukon	0.389	(0.313-0.476)	0.383	(0.304 - 0.460)	0.137	(0.076 - 0.192)		
Canada Border	0.091	(0.019 - 0.155)	0.053	(0.000-0.120)	0.081	(0.013 - 0.170)		
Pelly	0.169	(0.089 - 0.274)	0.076	(0.010 - 0.226)	0.265	(0.154 - 0.383)		
Tatchun	0.223	(0.113-0.291)	0.436	(0.295 - 0.509)	0.448	(0.315 - 0.538)		
Takhini	0.003	(0.000-0.032)	0.000	(0.000 - 0.048)	0.003	(0.000 - 0.048)		
Whitehorse	0.002	(0.000-0.041)	0.023	(0.000-0.067)	0.052	(0.000-0.105)		
Country								
US	0.512	(0.443 - 0.583)	0.413	(0.335 - 0.493)	0.152	(0.098 - 0.219)		
Canada	0.488	(0.417-0.557)	0.587	(0.507-0.665)	0.849	(0.781-0.902)		



Figure 1.—Locations of Chinook salmon populations from the Yukon River drainage surveyed for variation at SNP loci.

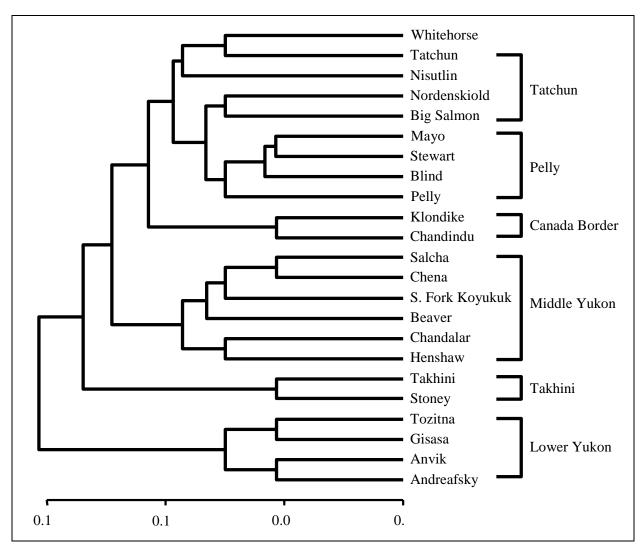


Figure 2.—Unweighted paired group-mean clustering tree based on genetic distances between pairs of Chinook salmon populations in the Yukon River drainage.

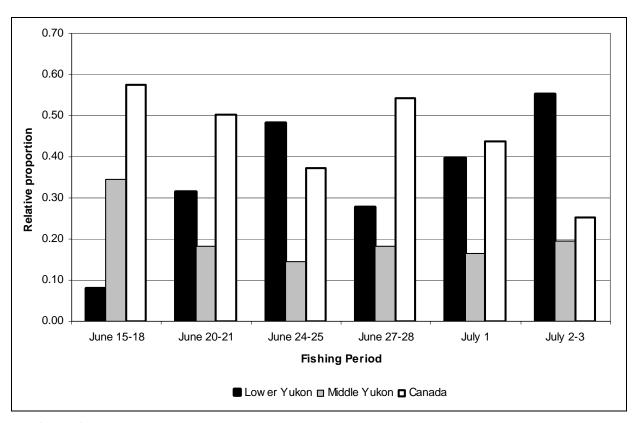


Figure 3.—Relative stock composition of three broad-scale reporting groups in the Chinook salmon harvest during the first six commercial fishery periods in District Y-1, 2004.

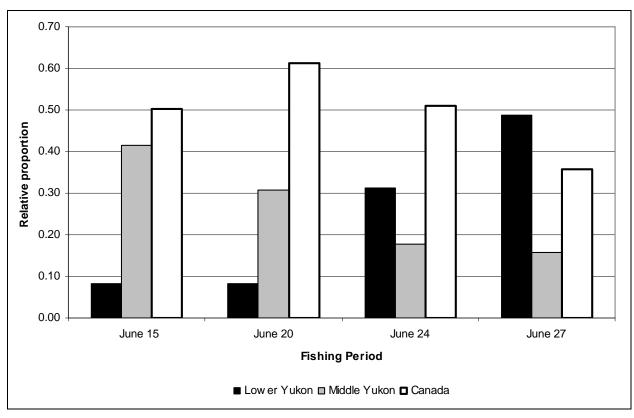


Figure 4.—Relative stock composition of three broad-scale reporting groups in the Chinook salmon harvest during the four commercial fishery periods in District Y-2, 2004.

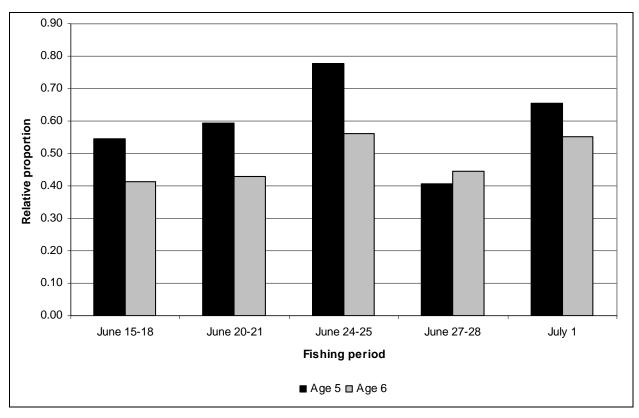


Figure 5.—Relative proportion of U.S. stocks in the five- and six-year old Chinook salmon harvested during the first six commercial fishery periods in District Y-1, 2004.

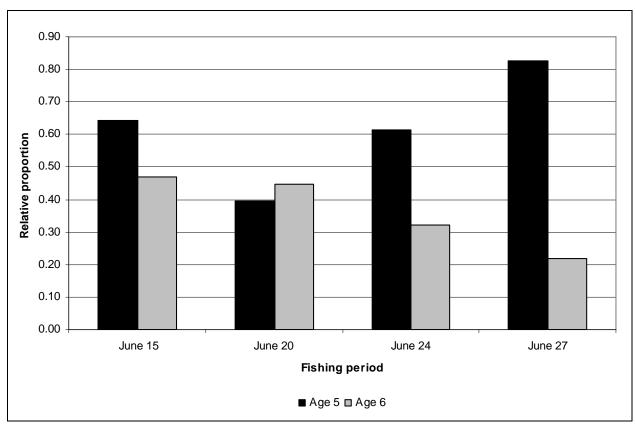


Figure 6.—Relative proportion of U.S. stocks in the five- and six-year old Chinook salmon harvested during the four commercial fishery periods in District Y-2, 2004.

APPENDIX A: ESTIMATED PROPORTIONAL STOCK COMPOSITION OF FIVE-YEAR OLD CHINOOK SALMON HARVESTED IN YUKON RIVER FISHERIES

Appendix A1.—Estimated proportional stock composition (P) and 90% confidence intervals of five-year old Chinook salmon harvested from the commercial fishery in District Y-1 of the Yukon River, 2004.

	Period 1 June 15-18		Period 2			Period 3 June 24-25		Period 4		Period 5	
			Jı	June 20-21				une 27-28	July 1		
		N = 58	N = 68			N = 73		N = 69	N = 68		
	P	90% CI	P	90% CI	P	90% CI	P	90% CI	P	90% CI	
Reporting Region											
Lower Yukon	0.127	(0.016 - 0.246)	0.506	(0.386-0.639)	0.667	(0.500 - 0.793)	0.338	(0.216 - 0.480)	0.534	(0.359-0.683)	
Middle Yukon	0.419	(0.266 - 0.594)	0.087	(0.025 - 0.212)	0.112	(0.007 - 0.262)	0.068	(0.003 - 0.156)	0.122	(0.000-0.273)	
Canada Border	0.063	(0.000 - 0.215)	0.140	(0.000-0.243)	0.000	(0.000 - 0.058)	0.061	(0.000 - 0.165)	0.075	(0.000-0.162)	
Pelly	0.201	(0.037 - 0.378)	0.000	(0.000-0.000)	0.040	(0.000 - 0.156)	0.000	(0.000 - 0.076)	0.032	(0.000 - 0.195)	
Tatchun	0.100	(0.000-0.260)	0.191	(0.076-0.311)	0.055	(0.000 - 0.179)	0.318	(0.131 - 0.464)	0.172	(0.042 - 0.297)	
Takhini	0.052	(0.000-0.143)	0.077	(0.000-0.180)	0.126	(0.017-0.217)	0.137	(0.018 - 0.270)	0.065	(0.000-0.167)	
Whitehorse	0.038	(0.000-0.128)	0.000	(0.000 - 0.044)	0.000	(0.000-0.000)	0.078	(0.000 - 0.209)	0.000	(0.000-0.000)	
Country											
US	0.546	(0.402 - 0.700)	0.593	(0.493-0.761)	0.779	(0.643 - 0.872)	0.407	(0.289 - 0.560)	0.656	(0.500 - 0.776)	
Canada	0.454	(0.300 - 0.598)	0.407	(0.239-0.507)	0.221	(0.128-0.357)	0.593	(0.440 - 0.711)	0.344	(0.224-0.500)	

Appendix A2.—Estimated proportional stock composition (P) and 90% confidence intervals of five-year old Chinook salmon harvested from the commercial fishery in District Y-2 of the Yukon River, 2004.

_		Period 1	Period 2			Period 3	Period 4		
	June 15 N = 65			June 20 N = 62		June 24	June 27 N = 89		
						N = 73			
	P	90% CI	P	90% CI	P	90% CI	P	90% CI	
Reporting Region									
Lower Yukon	0.219	(0.079 - 0.330)	0.070	(0.000-0.153)	0.479	(0.354 - 0.614)	0.663	(0.516 - 0.765)	
Middle Yukon	0.425	(0.245 - 0.595)	0.326	(0.176 - 0.464)	0.134	(0.026 - 0.239)	0.165	(0.075 - 0.270)	
Canada Border	0.152	(0.000-0.299)	0.020	(0.000-0.175)	0.016	(0.000-0.127)	0.025	(0.000-0.085)	
Pelly	0.000	(0.000-0.153)	0.129	(0.000-0.378)	0.087	(0.000-0.222)	0.032	(0.000-0.099)	
Tatchun	0.204	(0.055 - 0.349)	0.392	(0.160 - 0.554)	0.157	(0.013 - 0.273)	0.075	(0.000-0.164)	
Takhini	0.000	(0.000-0.042)	0.000	(0.000 - 0.047)	0.073	(0.000-0.204)	0.041	(0.000-0.122)	
Whitehorse	0.000	(0.000-0.035)	0.063	(0.000-0.152)	0.053	(0.000-0.158)	0.000	(0.000-0.098)	
Country									
US	0.644	(0.453 - 0.767)	0.395	(0.247-0.519)	0.613	(0.484 - 0.726)	0.827	(0.699 - 0.909)	
Canada	0.356	(0.233-0.547)	0.605	(0.481 - 0.753)	0.387	(0.274 - 0.516)	0.173	(0.091 - 0.302)	

Appendix A3.—Estimated proportional stock composition (P) and 90% confidence intervals of five-year old Chinook salmon harvested from the subsistence fisheries in districts Y-1 and Y-4 and the commercial fishery in District Y-5 of the Yukon River, 2004.

	D	istrict Y – 1	D	vistrict Y – 4	District $Y - 5$ $N = 119$		
		N = 119		N = 48			
	P	90% CI	P	90% CI	P	90% CI	
Reporting Region							
Lower Yukon	0.142	(0.044 - 0.254)	0.000	(0.000 - 0.064)	0.064	(0.000-0.166)	
Middle Yukon	0.407	(0.264 - 0.533)	0.397	(0.232 - 0.552)	0.137	(0.040 - 0.252)	
Canada Border	0.089	(0.000 - 0.218)	0.111	(0.000-0.250)	0.080	(0.000-0.194)	
Pelly	0.121	(0.017 - 0.284)	0.108	(0.000 - 0.287)	0.167	(0.022 - 0.356)	
Tatchun	0.145	(0.004 - 0.268)	0.384	(0.124 - 0.553)	0.487	(0.290 - 0.620)	
Takhini	0.058	(0.000 - 0.150)	0.000	(0.000 - 0.064)	0.004	(0.000 - 0.065)	
Whitehorse	0.038	(0.000-0.129)	0.000	(0.000-0.193)	0.061	(0.000-0.173)	
Country							
US	0.549	(0.415-0.665)	0.397	(0.238 - 0.558)	0.201	(0.090 - 0.323)	
Canada	0.451	(0.335-0.585)	0.603	(0.442-0.762)	0.799	(0.677-0.910)	

APPENDIX B: ESTIMATED PROPORTIONAL STOCK COMPOSITION OF SIX-YEAR OLD CHINOOK SALMON HARVESTED IN YUKON RIVER FISHERIES

Appendix B1.—Estimated proportional stock composition (P) and 90% confidence intervals of six-year old Chinook salmon harvested from the commercial fishery in District Y-1 of the Yukon River, 2004.

	Period 1 June 15-18 N = 293		Period 2			Period 3 June 24-25 N = 267		Period 4		Period 5	
			J	June 20-21 N = 265				une 27-28		July 1	
								N = 274		N = 258	
	P	90% CI	P	90% CI	P	90% CI	P	90% CI	P	90% CI	
Reporting Region											
Lower Yukon	0.075	(0.042 - 0.122)	0.224	(0.152 - 0.282)	0.384	(0.297 - 0.455)	0.218	(0.161 - 0.285)	0.368	(0.295 - 0.446)	
Middle Yukon	0.338	(0.270 - 0.411)	0.206	(0.147-0.301)	0.179	(0.115 - 0.262)	0.227	(0.164 - 0.290)	0.183	(0.122 - 0.271)	
Canada Border	0.071	(0.000-0.135)	0.016	(0.000 - 0.085)	0.000	(0.000 - 0.045)	0.034	(0.000 - 0.103)	0.000	(0.000 - 0.035)	
Pelly	0.121	(0.029 - 0.237)	0.157	(0.059 - 0.240)	0.104	(0.028 - 0.193)	0.033	(0.000 - 0.143)	0.187	(0.065 - 0.251)	
Tatchun	0.269	(0.171 - 0.351)	0.285	(0.169 - 0.365)	0.230	(0.130 - 0.330)	0.349	(0.226 - 0.420)	0.210	(0.115-0.301)	
Takhini	0.064	(0.016 - 0.107)	0.059	(0.014 - 0.127)	0.066	(0.006 - 0.122)	0.074	(0.016 - 0.138)	0.053	(0.000-0.125)	
Whitehorse	0.063	(0.000-0.124)	0.054	(0.000-0.127)	0.037	(0.000 - 0.096)	0.066	(0.000 - 0.140)	0.000	(0.000 - 0.037)	
Country											
US	0.413	(0.352 - 0.493)	0.429	(0.361 - 0.519)	0.563	(0.485-0.641)	0.444	(0.375-0.521)	0.551	(0.483 - 0.643)	
Canada	0.587	(0.507 - 0.648)	0.571	(0.481-0.639)	0.437	(0.359-0.515)	0.556	(0.479 - 0.625)	0.449	(0.358-0.517)	

Appendix B2.—Estimated proportional stock composition (P) and 90% confidence intervals of six-year old Chinook salmon harvested from the commercial fishery in District Y-2 of the Yukon River, 2004.

		Period 1		Period 2	_	Period 3	Period 4 June 27		
		June 15		June 20		June 24			
		N = 278	N = 287			N = 271	N = 252		
	P	90% CI	P	90% CI	P	90% CI	P	90% CI	
Reporting Region									
Lower Yukon	0.054	(0.007 - 0.117)	0.080	(0.030 - 0.128)	0.211	(0.134-0.280)	0.425	(0.344 - 0.494)	
Middle Yukon	0.404	(0.314-0.501)	0.306	(0.243 - 0.399)	0.232	(0.164 - 0.341)	0.153	(0.090 - 0.243)	
Canada Border	0.065	(0.000-0.151)	0.142	(0.049 - 0.213)	0.089	(0.018 - 0.158)	0.065	(0.000-0.120)	
Pelly	0.195	(0.079 - 0.316)	0.181	(0.078 - 0.300)	0.124	(0.029 - 0.205)	0.066	(0.003-0.187)	
Tatchun	0.261	(0.128-0.357)	0.276	(0.176 - 0.368)	0.328	(0.226 - 0.405)	0.214	(0.100 - 0.298)	
Takhini	0.021	(0.000-0.067)	0.016	(0.000 - 0.072)	0.015	(0.000 - 0.060)	0.021	(0.000 - 0.082)	
Whitehorse	0.000	(0.000-0.040)	0.000	(0.000 - 0.026)	0.000	(0.000 - 0.061)	0.058	(0.000-0.133)	
Country									
US	0.458	(0.378 - 0.550)	0.386	(0.321 - 0.465)	0.443	(0.373 - 0.522)	0.578	(0.503 - 0.663)	
Canada	0.542	(0.450 - 0.622)	0.614	(0.535 - 0.679)	0.557	(0.478 - 0.627)	0.422	(0.337 - 0.497)	

Appendix B3.—Estimated proportional stock composition (P) and 90% confidence intervals of six-year old Chinook salmon harvested from the subsistence fisheries in districts Y-1 and Y-4 and the commercial fishery in District Y-5 of the Yukon River, 2004.

	D	Pistrict Y – 1	D	istrict Y – 4	District Y – 5		
		N = 184		N = 179	N = 158		
	P	90% CI	P	90% CI	P	90% CI	
Reporting Region							
Lower Yukon	0.115	(0.036 - 0.179)	0.055	(0.003 - 0.118)	0.000	(0.000 - 0.038)	
Middle Yukon	0.344	(0.252 - 0.474)	0.379	(0.279 - 0.473)	0.162	(0.068 - 0.244)	
Canada Border	0.163	(0.054 - 0.254)	0.010	(0.000 - 0.101)	0.009	(0.000 - 0.159)	
Pelly	0.123	(0.023 - 0.294)	0.166	(0.040 - 0.323)	0.446	(0.230 - 0.576)	
Tatchun	0.255	(0.109 - 0.359)	0.353	(0.193 - 0.480)	0.271	(0.148 - 0.451)	
Takhini	0.000	(0.000-0.000)	0.019	(0.000 - 0.078)	0.056	(0.000-0.129)	
Whitehorse	0.000	(0.000-0.000)	0.017	(0.000-0.067)	0.056	(0.000-0.125)	
Country							
US	0.460	(0.371 - 0.562)	0.434	(0.334-0.521)	0.162	(0.076 - 0.255)	
Canada	0.540	(0.438-0.629)	0.566	(0.479-0.666)	0.838	(0.745-0.924)	